

FIRST ESTIMATE OF THE EGG PRODUCTION OF *ANGUILLICOLA CRASSUS* (NEMATODA: DRACUNCULOIDEA)

K. Thomas and F. Ollevier

Zoological Institute, Laboratory for Ecology and Aquaculture, Catholic University of Leuven, Naamsestraat 59, 3000 Leuven, Belgium

Investigating the life cycle of nematodes requires an estimate of the total number of eggs produced by a single female. No data are available concerning the fecundity of *Anguillicola crassus* (Kuwahara, Niimi et Itagaki, 1974). The objective of this study aimed at counting electronically the total number of eggs in the female reproductive organs at a certain time.

Live female *Anguillicola crassus*, recovered from swimbladders of eels, were weighed wet and dissected in Ringer-solution in watch glasses. All glassware was coated with silicone to prevent adhesion of eggs. After removal of intestinal and uterine tissues, eggs were fixed in 4% formaldehyde to prevent hatching. The sample was then diluted in a 0.9 % NaCl solution to a volume of 6 ml and split into two subsamples of 3 ml under continuous stirring. Each subsample was then adjusted to 100 ml with 0.9% NaCl and counted in an electronic counter, Coulter (R) Multisizer AccuComp (R) Rev. 1.10 (Coulter Electronics Ltd.). The 200-µm pore size was judged appropriate to count *A. crassus* eggs as spherical objects (the electronic counter only counts spherical particles). For each 100 ml subsample 5 successive counts of 2 ml were performed resulting in two estimates of the egg content of the worm. This procedure was followed for three adult female worms: (1) a non-fertilized female, (2) a fertilized female which had not yet shed eggs and (3) a fertilized female which had shed already several thousands of eggs. The third female was kept alive in Ringer-solution during three days and the number of eggs shed daily was counted.

Eggs are slightly elliptical and width varied between 50.2 and 88.3 µm (mean: 69.9 µm; n = 72) and length between 53.2 and 119.8 µm (mean: 86.0 µm) for uterine eggs for various developmental stages. The size of the eggs as measured by the Coulter Multisizer AccuComp did not coincide with the real values, but were always smaller. This is probably caused by the non-interruption of the electric current by the eggshell; the diameter of the particle is estimated from the egg core or larvae in the egg. The core of the eggs measured between 38.8 (mean width) and 48.3 µm (mean length) and mean value of the diameter measured by the Coulter Multisizer was 43.3 µm.

The estimated total number of eggs for each *A. crassus* is given in Table 1. The three worms differ in their absolute egg content, but the results are indicative for the total number stored at a time in a female: 100 000 - 150 000. The first female was not yet fertilized and had the lowest egg content, the second one was fertilized and had the highest egg content (no eggs in the swimbladder lumen). The third had a lower egg content than the

previous one but at least 46 000 were already deposited, of which half was laid in just three days.

Table 1. *Anguillicola crassus*: A) Egg content of adult females and B) eggs deposited by a female. Number of eggs are counted with the Coulter Multisizer AccuComp, except for the egg output of female 3.

	Wet weight (mg)	No. of eggs / subsample	Estimated total / female
A) Female 1	165	4 501	90 020
Female 2	221	5 208	104 160
		8 214	164 280
Female 3	>220	8 383	167 660
		7 013	140 260
		7 780	155 600
B) Female 3:		eggs in lumen:	21 800
		eggs deposited after 24h:	2 410
		eggs deposited after 48h:	9 350
		eggs deposited after 72h:	12 600
		total egg output:	46 160

The use of a Coulter counter for counting nematode eggs was proposed by Smith (Smith J. W. 1987: Can. J. Zool. 66: 2253-2254). Although in this study a different electronic counter (Coulter Multisizer) was used, the method is appropriate for estimating the total egg content, but not for sizing the eggs. We obtained a first estimate of the egg content of *Anguillicola crassus*, although many factors which influence egg production could not be taken into consideration. The egg content of *Contracaecum osculatum* varied between 5 000 and 33 000 and of *Hysterothylacium aduncum* between 8 000 and 23 000 for nematodes of various weight classes (Smith 1987, op. cit.). For two more related nematodes within the order of the Dracunculoidea, egg content was estimated on 75 000 for a 4 cm long *Philometra ovata* (Moravec F. 1980: Folia Parasitol 27: 29-37) and 40 500 for a 9 cm long, and 100 000 for a 17 cm long *P. obturans* (Moravec F. 1978: Folia Parasitol. 25: 303-315). Moravec (1978, 1980; op. cit.), however, did not use an electronic counter. The high fecundity of *A. crassus* has certainly facilitated the rapid spread of this parasite in the European eel population (e. g., Belpaire C., De Charleroy D., Grisez L., Ollevier F. 1989: Eur. Int. Fish. Adv. Comm. (FAO), Working Party on Eel, Porto, Portugal, May 29 - June 3; Kennedy C. R., Fitch D. J. 1990: J. Fish Biol. 36: 117-131).

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